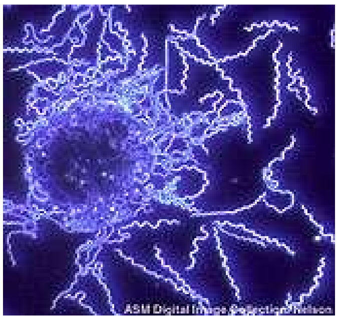

Microbes: Transformers of Matter and Material

"Microbes can do anything they want, wherever they want - without microbes, humans wouldn't be alive"

In this lesson, we wish to ask:

- What is the diversity of microbes?
- How do the different kinds of microbes function in gaining energy?
- What impacts do microbes have on ecosystems and our globe?

Points to Remember:

- Microbes can do anything they want, wherever they want.
- Without microbes, humans wouldn't be alive.

Definition: Microbes

Microbes are organisms that we need a microscope to see. The lower limit of our eye's resolution is about 0.1 to 0.2 mm or 100 - 200 um. Most microbes range in size from about 0.2 um to the 200 um upper limit, although some fruiting bodies of fungi can become much larger. Microbes include the bacteria, algae, fungi, and protozoa. In this lecture we will discuss mostly the bacteria and the fungi.

Evolution

There are two major groups of bacteria, the "eubacteria" and the recently discovered "archaebacteria". The eubacteria contain most of the common bacteria such as *E. coli* and the cyanobacteria (blue-green algae). The archaebacteria are found mainly in the deep ocean near hydrothermal vents. What is striking from the standpoint of the divergence of genetic material (the order and sequences of genes), is that these two groups of bacteria are more different than are animals and plants. In other words, these two groups of bacteria have evolutionarily diverged further from one another than animals have diverged from plants.

Introduction to Some of the Important Microbes

(A) Bacteria

Bacteria are found everywhere in water, soil, and even air. These small prokaryotic cells, typically from 0.2 to 1 um in length, are capable of living in boiling water, frozen ground, acid volcanoes, and at the bottom of the ocean (for a refresher on the different kinds of "cells",). They can reproduce by doubling with a generation time of 20 minutes, or survive for centuries in a resting stage. In natural waters (lakes, streams, oceans) their generation time is around 1 day. In soils they live in a film of water around plant roots or other particles, and their activity is dependent on the temperature and the amount of available moisture. In general, bacteria are found in concentrations of 10^6 cells/mL of water in surface waters, and 10^9 cells/mL of soil in soils and sediments.

Some bacteria are capable of locomotion, and they possess the only rotary motor known in all of biology. This motor, similar to a wheel and axle, is capable of spinning a flagellum at speeds of 100 revolutions per second, or 6,000 rpm. Bacteria can propel themselves at a rate of 10 times their body length each second.

Bacteria, like all cells, are composed mostly of carbon, oxygen, nitrogen, hydrogen, phosphorus, and sulfur in the following percentages:

Element	***% of dry weight***
C	55
O	20
N	10
H	8
P	3
S	1

Bacteria take these elements and arrange them into polymers in the cells in the following percentages:

52.4% protein (amino acids, CHNOS)

19.9% nucleic acid (organic bases, CNOHP)

16.6% polysaccharide (sugar, CHO)

9.4% phospholipid (C-16 acid + P, CHOP).

(B) Fungi

Fungi grow in the form of a finely-branched network of strands called hyphae which are 5-10 um in diameter. These hyphae can release digestive enzymes and take up nutrients over their entire length. Fungi can absorb only small molecules such as sugars or peptides less than size amino acids. The reproductive organs of the fungi are called fruiting bodies or sporangia, which are sacs or other tissues that contain the fungi spores.

Fungi are uncommon in aquatic environments. On land, the amount of hyphae in the soil is measured in hundreds or thousands of meters of length per gram of soil. For example, the total

length of hyphae in a gram of soil (about the amount that would fit on the fingernail of your little finger) can reach up to 1,600 meters (think about that for a minute).

Fungi secrete enzymes that can break down cellulose into glucose, one of the few kinds of organisms able to do this. Fungi are the only known organisms that degrade lignin completely. Cellulose and lignin are structural materials in plants that are difficult to degrade. The fungi do not use the breakdown products of lignin, but instead they use hydrogen peroxide to oxidize lignin in place. The breakdown products diffuse away, exposing the cellulose to enzymatic attack.

(C) Protozoa

Protozoans are single-celled eukaryotes, not photosynthetic, that move by flagella or cilia. In oceans and lakes, the small 2-10 um long flagellates are the most important predators on bacteria. The larger ciliates (e.g., *Paramecium*) prey mostly upon photosynthetic cyanobacteria and small eukaryotic algae. In some termites, anaerobic protozoans in the gut degrade cellulose.

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4. How do Bacteria Gain Energy to Grow?

** Assimilative versus Dissimilative processes*

Microbes must acquire certain elements to grow and reproduce -- these elements compose their protoplasm in the proportions listed in the table above. In addition, they must produce ATP in order to use the stored energy in this molecule to operate various cellular processes. *Assimilative processes* are used to bring needed elements into the cell and to incorporate them into the cell protoplasm. *Dissimilative processes* do not incorporate elements into the cell, but instead they use the energy gained in the process to form ATP.

Microorganisms are classified as autotrophs or heterotrophs based on whether or not they require pre-formed organic matter. Autotrophs derive energy from either light absorption (photoautotrophs) or oxidation of inorganic molecules (chemoautotrophs). In most of the light reactions the bacteria are fixing carbon dioxide into organic carbon, just as green plants do. Some photosynthetic bacteria (photoheterotrophs) require pre-formed organic matter as reducing agents, but generate ATP from the absorption of light energy. Finally, some bacteria and fungi (heterotrophs) used pre-formed organic matter as both a source of energy to generate ATP and as a source of carbon for the cell, just as animals do. The following table summarizes the classification of the ways in which microbes process energy.

Classification	***Energy source for generating ATP***	***Source of carbon for the cell***	***Example of organisms***
Photoautotroph	Light	CO_2	Bacteria, plants
[illegible]	[illegible]	[illegible]	[illegible]

	compounds		
Photoheterotroph	Light	CO_2, organic matter	Bacteria
Heterotroph	Organic matter	Organic matter	Bacteria, fungi, animals

As an example of the diversity of dissimilatory reactions that bacteria use to produce energy, consider the following table that shows various reduction-oxidation reactions (for a primer on "redox" reactions, please click here). Note that all of these reactions listed below are performed by chemoautotrophs. A "+" in the table indicates that bacteria can use the pair of electron acceptor and donor to run a redox reaction that produces sufficient energy for growth. A "-" indicates that bacteria cannot use the redox pair for growth (this is not a table to memorize). CHO is a shorthand for organic matter containing Carbon, Hydrogen, and Oxygen.

Reductant (right)	H_2	CHO	CH_4	HS^-
Oxidant or electron acceptor *(below)*				
CHO	+	+	-	-
CO_2	+	+	-	-
SO_4^{2-}	+	+	?	-
NO_3^-	+	+	?	+
O_2	+	+	+	+

Some of the common dissimilative reactions that bacteria perform to gain energy and to decompose organic matter are listed in detail in the table below (note that you can match-up the redox pairs in the table above to the full reactions in the table below). The energy yield listed in the last column is the amount of energy in kilocalories that is produced per mole of oxidant that is used. The formation of ATP requires about 7 kcal/mol of energy, so only reactions producing more than 7 kcal/mol can be used by bacteria for growth. As the most powerful oxidants (the electron donors that generate the greatest energy yields) are consumed, the major reaction that is performed by the bacteria shifts to the next most energy yielding process. For example, when oxygen is depleted from the environment, nitrate reduction occurs. If oxygen becomes available again, then nitrate reduction will stop, even if there is still NO_3^- available in the environment, and aerobic respiration will continue. This shifting of electron donors continues until only CO_2 is left to serve as an oxidant, in which case the bacteria reduce the CO_2 to methane, CH_4. It is interesting to note that some bacteria can perform more than one of these reactions, whereas other bacteria are quite specialized and can perform only a

single, specific dissimilatory reaction. Remember that in all of these reactions the bacteria must have a source of carbon to incorporate into their cells in order to grow.

Reaction name	***Reductant***	***Oxidant***	***Reaction Stoichiometry***	***Energy Yield (kcal/mol)***
Aerobic Respiration	CHO	O_2	$C_6H_{12}O_6 + 6O_2 <==> 6CO_2 + 6H_2O$	686
Nitrate Reduction	CHO	NO_3^-	$CHO + NO_3^- + H^+ <==> CO_2 + N_2 + H_2O$	649
Sulfate Reduction	CHO	SO_4^{2-}	$2CHO + SO_4^{2-} + 2H^+ <==> 2CO_2 + HS^- + 2H_2O$	190
Methanogenesis	H_2	CO_2	$4H_2 + CO_2 <==> CH_4 + 2H_2O$	8.3

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5. What are the Important Impacts of Microbes on Ecosystems?

(1) Generate Oxygen in the Atmosphere.

Almost all of the production of oxygen by bacteria on earth today occurs in the oceans by the cyanobacteria or "blue-green algae.

(2) Recycle nutrients stored in organic matter to an inorganic form.

Decomposition releases the mineral nutrients (e.g., N, P, K) bound up in dead organic matter in an inorganic form that is available for primary producers to use. Without this recycling of inorganic nutrients, primary productivity on the globe would stop.

On land, most of the decomposition (also called "mineralization") of dead organic matter occurs at the soil surface, and the rate of decomposition is a function of moisture and temperature (too little or too much of either reduces the rate of decomposition). Fungi are important in terrestrial systems, but not in aquatic. They are present even before the leaves and twigs enter the soil and so decomposition starts in the living or senescent plant material. Fungi are the most important decomposers of structural plant compounds (cellulose and lignin – but note that lignin is not broken down when oxygen is absent). The fungi invade the organic matter in soils first and are then followed by bacteria.

In water, the decomposition of organic matter is mostly oxic in streams and in the ocean and anoxic in the bottoms of lakes or in swamps. As shown in the table above, oxic decomposition proceeds faster (produces higher energy yields for the bacteria) than decomposition in environments where there is no oxygen. In the open ocean, the water is so deep (average 3900

m) and contains so much oxygen, that most of the algal-formed organic matter at the surface decomposes aerobically before it reaches the bottom. For example, only 2% of the primary productivity in the upper ocean sinks to a depth of 3500 m. Most of the world is ocean, and most of the ocean is deep, so most of the aquatic decomposition must be aerobic. But in shallow waters, coastal oceans, lakes and estuaries, 25-60% of the organic matter produced may settle out of the upper waters rapidly and be decomposed anaerobically.

Of course another important impact of decomposition besides generating inorganic nutrients is to produce CO_2 and CH_4 that is released to the atmosphere.

(3) Fix nitrogen from the Atmosphere into a Useable Form.

The only organisms capable of removing N_2 gas from the atmosphere and "fixing" it into a useable nitrogen form (NH_3) are bacteria. The specific bacteria that can perform N fixation are scattered throughout the groups including the cyanobacteria. All organisms that fix nitrogen use the same mechanisms and the same enzymes – this ability probably evolved only once and early in the history of life. Symbiotic N_2 fixation costs the plant photosynthate to support the fixation and the NH3 assimilation; this cost could be from 15-30% of the total carbon assimilated by the plant. In fact, to fix one molecule of N_2 requires about 25 molecules of ATP, so it is expensive from the bacterial standpoint, and that means that the plant must support that energy requirement. In return the plant receives nitrogen, which may otherwise be a limiting nutrient.

Another difficulty for the bacteria is that one of the enzymes necessary for nitrogen fixation is destroyed by oxygen (which is necessary for efficient ATP formation). One solution to this problem is to form symbiotic relationships with other organisms that can provide carbohydrates; these include diatoms, the fungi of certain lichens, shipworms, termites, and certain plants especially in nodules of the roots.

(4) Allow Herbivores to Consume Poor Quality Food.

In the ocean, most of the primary productivity is consumed by herbivores. In contrast, in terrestrial systems most of the primary productivity is not consumed by the herbivores. The reasons for this difference are: (1) animals lack digestive enzymes capable of using cellulose and lignin and other structural plant compounds; (2) plants often have anti-grazing toxins, aromatic resins, or thorns; (3) most land plant tissue is poor in mineral nutrients (especially N and P) compared to the tissue in the herbivore.

Plant Community	% of primary production consumed by herbivores
Phytoplankton (open water)	60 - 90
Grasslands	12 - 45
Kelp beds	10
Salt marshes	7

Mangroves	5
Deciduous forests	1.5 - 5

In a ruminant animal (cattle, deer, giraffe) the ingested food, possibly regurgitated and re-chewed, passes into the rumen together with saliva (60-100 liters produced per day). The rumen is really a continuous fermenter where the complex carbohydrates of the plant are fermented into methane, carbon dioxide, and fatty acids. The biota of the rumen are found in about equal biomasses of bacteria (10^{11}/ mL), protozoans (10^5/ mL to 10^6/ mL), and fungi (poorly known biomass). About 60-65% of the total energy removed from the plant food that is ingested by the animal comes from rumen fermentation. Plant tissues passing from the rumen undergo secondary fermentation in the caecum and large intestine where an additional 8-30% of the total energy is provided.

In addition, many termites contain protozoans and bacteria in their guts that perform similar operations. The protozoans are capable of digesting cellulose, and bacteria in the gut generate CH_4 from the organic compounds released from the cellulose degradation. Finally, some termites also have bacteria in their guts that are capable of fixing nitrogen from the atmosphere, providing a useable nitrogen source for the termite.

(5) Give Plant Roots Access to Nutrients in the Soil.

Plant roots create a zone of nutrient depletion around themselves. To have access to new sources of nutrients, a plant can either grow more roots and small root hairs (some as small as 10 um) or form an association with a fungus whose hyphae provide an even more efficient absorptive structure. Most vascular plants can form such associations, which are called "mycorrhizae". Mycorrhizal fungi include those living on the surface of plants (ectotrophic or sheathing) and those which enter the host (endotrophic or vesicular-arbuscular or simply "V-A").

The added advantage to the plant is that the hyphae can secrete enzymes that break down organic molecules and make inorganic nutrients available. While the plants gain nutrients, the fungi gain carbohydrate food from the plant. There is also a cost to the plant in this association; one study reported that mycorrhizal biomass was only 1% of a fir forest ecosystem but used 15% of the net primary production.